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Review on *Parthenium hysterphorus* as a potential energy source

Ghosh Swati [a], Haldar S. [b], Ganguly A. [c], Chatterjee P.K. [c,*]

[a] Department of Chemistry, Institute of Engineering and Industrial Technology, Durgapur 713212, India
[b] Department of Chemical Engineering, National Institute of Technology, Durgapur 713209, India
[c] Thermal Engineering Group, Central Mechanical Engineering Research Institute, Durgapur 713209, India

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ABSTRACT

Ethyl alcohol, the oldest synthetic organic chemical is rendered one of the most important alternative sources of energy. Efficient production of ethanol is based on optimized processes where utilization of cheap substrates is highly demanding. Utilization of different types of lignocellulosic materials can be considered for production of ethanol. Amongst the various types of lignocellulosic substances Parthenium [*Parthenium hysterophorus* L. (Asteraceae)] is a potential resource available in many tropical regions of the world. A considerable amount of laboratory work is in progress for bioconversion of various lignocellulosic materials into ethanol using sequential steps of hydrolysis, saccharification and fermentation. However, there is very little reported work for bioconversion of *P. hysterophorus* into fuel ethanol. A comprehensive review on the availability of Parthenium, its composition and impact on human and other livestock, bioconversion into ethanol, the methods of pretreatment and the chemistry of hydrolysis with dilute sulfuric acid has been attempted in this work. The results of depolymerization for a hypothetical pentamer can be predicted through solution of the governing equations through simple integration technique. The series of reactions interplay during the depolymerization of hemicellulose. The same can be successfully used to predict the yield of xylose when parthenium feedstock is hydrolyzed with dilute sulfuric acid. The present report will stimulate the researchers to adopt a suitable kinetic model to study the reaction mechanism for hydrolysis of *P. hystrophorus* L. and optimization of different parameters such as temperature, time, acid concentrations, alkali concentrations, etc. during pretreatment processes for achieving higher yield of ethanol.

Contents

* Corresponding author, Tel.: +91 343 6510236; fax: +91 343 2546745.
E-mail addresses: pradipcmeri@gmail.com,
pradip@cmeri.res.in (P.K. Chatterjee).

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1. Introduction

The energy crisis caused through the depletion of traditional fossil fuel sources such as coal and petroleum is an unquestionable issue of today's world. During last few years it has been essential to accept biofuels like ethyl alcohol as a viable potential substitute for the petroleum. Elevating problems regarding global warming and crude oil prices over recent years reignited public interest in fuels and chemicals derived from renewable sources like lignocellulosic materials [1]. In addition, global economic downturn offers an opportunity to cultivate the green technology while costs are lower and green growth is the only desired future for growth and progress of human society. Lignocellulosic materials being cheap, renewable and eco-friendly in nature, is a potential feedstock for the production of second-generation biofuels. Sources of lignocellulosic biomass include agricultural residues, woody biomass, or some wasteland weeds. It is more abundant and less expensive than food crops like sugar, starch etc. for ethanol generation [2].

Lignocellulosic biomass is composed of cellulose, hemicelluloses and lignin. Cellulose and hemicellulose include the carbohydrate portion of biomass and lignin consists of poly (aromatic) moieties from phenylpropanoid building blocks. Minor constituents are organic extractives (mainly terpenes) and inorganic compounds (ash). Current biofuel productions through green technologies focus on the utilization of the carbohydrate fraction, for ethanol production [3]. *Parthenium hysterophorus* being a lignocellulosic biomass may be considered as the source of ethanol. It is an annual asteraceous herb and one of the most obnoxious weed in the world. This weed is considered to be a cause of allergic respiratory problems, contact dermatitis, mutagenicity in human and livestock. Crop production is drastically reduced owing to its allelopathy. Also aggressive growth of this weed threatens biodiversity. It is native to Central and South America and is considered to have originated from the Gulf of Mexico. It has spread rapidly and extensively throughout the world since 1970 and is considered a major threat in many regions.

P. hysterophorus is able to grow on a wide range of soil types ranging from sandy to heavy clays but growth is better in later type of soil. It occurs in areas with summer rainfall greater than 500 mm per annum. Germination favours at temperatures between 10 °C and 25 °C. Parthenium weed colonises new areas rapidly by means of relatively high numbers of seeds, producing one lakh seeds per plant. Dispersal of seed takes place via vehicle, water, animals, farm machinery and wind. Disturbed habitats such as roadsides and railway tracks, stockyards, buildings surroundings and fallow agricultural land are particularly suitable for growing *P. hysterophorus* due to a lack of interspecies competition [4]. Being a member of the Asteraceae family, *P. hysterophorus* can be confused with a number of other introduced weed from the family including seedling of cobbler's pegs (*Bidens subaltermans*), flowers of bishop's weed (*Ammi majus*) and hemlock (*Conium macultum*), and the ragweeds (*Ambrosia* spp.) [5].

1.1. The biology of P. hysterophorus: botanical description and germination of parthenium

Parthenium, as shown in Fig. 1, is an annual herbaceous member of the Asteraceae with a deep tap root and an erect stem that gradually changes into semi-woody with age. It branches itself out usually up to about 1–2 m. It has bi-pinnatfied and pale green leaves covered with soft fine hairs. Parthenium can grow and reproduce itself any time of the year. During a favorable growing season, four or five successive generations of seedlings can emerge at the same site. The photosynthetic characteristic of parthenium leaf is mostly related to C_3 type pathway and exhibits a photosynthesis rate of 25–35 °C and a high CO_2 level. Low temperature (< 10 °C) considerably reduces plant growth, mainly flowering and seed production by reducing leaf area index, relative growth rate and net assimilation rate and leaf area duration. The weed grows fast and comfortably on alkaline to neutral clay soils. However, its growth is slow and less prolific on a wide range of other soil types. Parthenium is a prolific seed producer. For example, in a highly infested field in India, a single plant produced 2,00,000 seeds/m^2. The germination process of the weed involves several steps required to change the quiescent embryo to metabolically active embryo. For a seed to germinate adequate water, suitable temperature and composition of gases (O_2/CO_2 ratio) in the atmosphere and light should be available [6]. Several aspects of the ecology of parthenium weed contribute to its aggressiveness. These includes size and persistence of the soil seed bank, longevity of seed when buried, fast germination rate and the innate dormancy mechanism of seed which makes it well adapted to semi-arid environments and increases the chances of seed burial.

Since *P. hysterophorus* does not reproduce vegetatively from plant parts, the only method of reproduction and spread is by seed. Seed spread by wind is limited. Movement in sheet water flow is important as indicated by large colonies along waterways and on drainage floodplains. Most long distance dispersal of seed is by vehicles and farm machinery as evidenced by major spread of parthenium along roads. A period of drought followed by rain provides ideal conditions for spread. Drought reduces pasture cover and increased movement of stock and stock fodder also aids the spread of seed. In particular, flooding after drought is advantageous to the weed as flood is a dispersal mechanism for *P. hysterophorus*.

In summer, plants can flower and set seed 4 weeks after germination because they are stressed and small. Buried seeds have been found to last much longer than seed on the soil surface. Timing of chemical control is critical so that parthenium weed is



Fig. 1. Photograph of *Parthenium Hysterophorus* L.

removed when plants are small and have not produced seed, and when grasses are actively growing and seeding to decolonize the infested area (e.g. in early summer). Studies suggest that after 6 years, 50% of seed buried 5 cm below the surface are still viable. However, unlike other weed species, there is no critical point where invention is required, as *P. hysterophorus* can produce flowers and seed at any time of the year under favorable conditions [5].

1.2. Geographical distribution

The wasteland weed *P. hysterophorus* has been recorded growing naturally since centuries in Mexico, Cuba, North and South America, West Indies, Australia, Taiwan, Southern China, Pacific island, East and South Africa and Canada. Till 1977 the weed did not find any place in the list of world's worst weeds. But within the last decade, it has become one of the seven most dreaded weeds of the world. In India, it is noticed only from mid-1950s and is presumed to have been accidentally introduced in Maharashtra. However, its spread all over the country has been rapid with abnormal density [7]. Parthenium weed was first noted in India near Poona in Maharashtra State in 1951. By 1972 it had spread into the majority of the states from Kashmir in the north to Kerala in the south. Continuing to spread, it was found in Assam in 1979 and is now present almost throughout the subcontinent and is probably the dominant weed in Karnataka State [8]. It has invaded nearly 4.25 million hectares of land in India.

1.3. Impact of parthenium

P. hysterophorus is an aggressive noxious annual herbaceous weed with no economic importance till now [9]. The rapid growth of the *P. hysterophorus* weed had become a threat to the environment and biodiversity. It adversely affects the germination and growth of several crops as well as human being and livestocks.

1.3.1. Human being

P. hysterophorus is a weed of global significance, occurring as an alien invader in over 20 countries in Africa, Asia and Oceania. It causes human health problems such as asthma, bronchitis, dermatitis and hay fever [6]. The contact dermatitis due to this has long been recognized. There are evidences of allergic papules in school boys when they had volunteered for uprooting parthenium in Hassan. It has been observed that chances of getting sensitized to the weed are high when a person comes in contact with the weed for a period ranging from 3 to 12 months. Parthenium is responsible for causing a largest number of airborne contact dermatitis in India. Almost every part of the plant except root is reactive. The reaction was mainly over the sun exposed area [7]. The homesteads of many rural communities are surrounded by dense *P. hysterophorus* infestations, resulting in continuous direct exposure to the weed. It is stipulated that allergic reactions to *P. hysterophorus* may be exacerbated in persons that are immune-compromised by diseases such as HIV and tuberculosis.

1.3.2. Livestock

Though cattle do not eat parthenium, its effect was observed on them when they walk by or graze through patches of this weed. Such cattle had inflamed under and subsequently suffered from fever and rashes. It is reported that feeding the weed to buffalo and bull calves at different level causes both acute chronic forms of toxicity. Ulcerations are caused both in the mouth and digestive track. An autopsy of the dead animals showed punched cut ulcers on the esophagus and the obosomal folds. Histopathology of the kidney and liver revealed degenerative changes and necrosis. It was reported that "Labeled parthenium" was found to be excreted in the milk when administered to lactating guinea pigs, rabbits and cow. Consumption of milk from the livestock grazing around parthenium invaded freezing land could be hazardous to human being [7].

1.3.3. Agriculture

The adverse impacts of *P. hysterophorus* on agriculture have been reviewed by several authors. In India, *P. hysterophorus* causes yield decline of up to 40% in agricultural crops. It reduces the pasture carrying capacity by up to 90% [7]. On cracking clay soils with an annual rainfall between 600 and 800 mm, *P. hysterophorus* is estimated to reduce the carrying capacity of affected farms in Australia by about 40%.

1.3.4. Effect of pollen

The pollen of this weed causes skin allergy and other allergic reaction such as asthma, cold reddening of eyes, inflammation of eyebrows etc. A few studies carried out in New Delhi revealed that parthenium pollens were observed in atmosphere throughout the year and that the pollen of parthenium showed marked positive skin reactions (2+ and 3+) in 14 out of 50 patients. They observed that flower extracts stand next to pollen in allergenicity [7]. The flower extracts showed positive skin reactions in 3 out of 50 patients (6%), closely followed by leaf extract (2 out of 50). The pollen also inhibits fruit set in crops such as tomato, brinjal, beans and capsicum and reduces grain filling in cereals such as maize and sorghum.

1.4. Chemical composition of P. hysterophorus

The chemical analysis of *P. hysterophorus* has indicated that all the plants parts including trichromes and pollen contain toxins called glycoside perthenin, major sesquiterpene lactones. Other phytotoxic compounds or allelochemicals are hysterin, ambrosin, flavonoids such as quercelagetin 3, 7-dimethylether, 6-hydroxyl kaempferol 3–0 arabinoglucoside, fumaric acid, *p*-courmeric, caffeic acid, vanilic acid, ansic acid, *p*-ansic acid, chlorogenic acid, ferulic acid, sitosterol and some unidentified alcohols. Parahydroxy benzoic acid is lethal to human beings and animals. Isolation and structural elucidation of the active principles of *P. hysterophorus* is required to determine their chemical properties. Parthenin, hymenin and ambrosin are found to be the culprits behind the hysternin, menacing role of this weed in provoking health hazards. *P. hysterophorus* from different geographical regions exhibited parthenin, hymenin, coronopilin, dihydroisoparthenin, hysternin, hysterophorin and tetraneurin as the principal constituents of their sesquiterpene lactones. A novel hydrooxyproline-rich glycoprotein has been identified as the major allergen in *P. hysterophorus* pollen. Latter the flowers of *P. hysteroporus* were examined and four acetylated pseudoguaianolides along with several known constituents were isolated. Sesquiterpenoid, charminarone, the first pseudoguaianolides, ambrosanolides have been found along with several known compounds from the whole plant [9]. The chemical and biological composition of parthenium in various form are shown in Tables 1–4.

1.4.1. Cell wall composition of P. hysterophorus

Cell wall of *P. hysterophorus* is mainly composed of cellulose, hemicellulose and lignin. Composition of various lignocellulosic biomasses [12] is shown in Table 5. The average biological composition of *P. hysterophorus* as measured in the laboratory are Lignin: 13.9; Cellulose: 27.8% and Hemicellulose: 21.01%.

Cellulose is the main constituent of *P. hysterophorus*. Approximately 40–45% of dry substance in most wood species is cellulose located predominantly in the secondary cell wall. Cellulose molecules are completely linear and have a strong tendency to form intra-molecular and intermolecular hydrogen bonds. Thus, bundles of cellulose molecules are aggregated together to form microfibrils in which highly ordered (crystalline) regions alternate with less ordered (amorphous) regions. Microfibrils build up fibrils and finally cellulose fibers [13]. The free hydroxyl groups present in the cellulose macromolecule are likely to be involved in a number of intra and intermolecular hydrogen bonds which may give rise to various ordered crystalline arrangements [14]. Cellulose chains are formed into microfibrils which constitutes the basic framework of the cell, conveying a great resistance to tensile forces [15]. Cellulose is a homopolysaccharide composed of β-D-glucopyranose units which are linked together by β–(1→4) glycosidic linkages. Fig. 2 shows that two adjacent glucose units are linked by elimination of one molecule of water between their hydroxylic groups at carbon 1 and carbon 4. The cellobiose unit is the repeating unit of the cellulose chain with a length of 1.03 nm [16]. Four principal allomorphs have been identified for

Table 1
Chemical and biological characteristics of composted parthenium [10].

Characteristics	Values
Macronutrients (%)	
Total N	1.58
Total P	0.33
Total K	1.64
Total S	0.29
Micronutrients (ppm)	
Fe	7829
Mn	304
Zn	116
Cu	66
Electrochemical	
pH	7.8
EC (dS m^{-1})	1
Biological (g $compost^{-1}$)	
Total bacteria	13.66×10^6
Fungi	9.67×10^4
Azotobacter	2.33×10^6
Actinomycetes	7.67×10^5
Phosphate solubilising bacteria (PSB)	2.67×10^6

Table 2
Parthenium ash and proximate analyses [11].

pH	10.74
Conductivity (S/cm)	1426
Particle size (B.S.S.)	150
Specific gravity	0.828
Water absorption (%)	31.292
Bulk density (g cm^{-3})	1.47
Apparent porosity (%)	46
Matter insoluble in water (%)	9.086
Surface area (m^2 g^{-1})	760.56
Moisture (%)	0.99
Volatile matter (%)	15.85
Ash content (%)	11.6
Fixed carbon (%)	72.55
Silica (ppm)	574
Iron (ppm)	11.5
Sodium (ppm)	7.03
Potassium (ppm)	3.39
Chromium	Not detectable
Nickel	Not detectable

Table 3
Infrared bands of parthenium ash along with their probable assignment [11].

Band Position (cm^{-1})	Assignment
3403	–OH stretching
2925, 1426	C–H stretching
1780, 1460	–C=O stretching
1116	Si–O stretching
1057	Al–O
892	Fe–O
691, 614	Ca–O
441	Na–O

Table 4
d-values of parthenium ash (x-ray defraction pattern) [11].

d (Å)	Possible component
4.30	Iron oxide (tetragonal)
2.65	Iron lead oxide (hexagonal)
3.53	Lead oxide (tetragonal)
2.90	Lead oxide (tetragonal)
1.82	Lead oxide (tetragonal)
1.54	Lead oxide (tetragonal)
1.28	Lead oxide (tetragonal)
3.37	Silicon oxide (hexagonal)
3.21	Silicon oxide (hexagonal)
3.17	Silicon oxide (hexagonal)
3.05	Silicon oxide (hexagonal)
3.02	Silicon oxide (hexagonal)
2.11	Silicon oxide (hexagonal)
1.41	Silicon oxide (hexagonal)
2.29	Silicon oxide(hexagonal)
2.23	Calcium manganese oxide (orthorhombic)
2.17	Calcium manganese oxide (orthorhombic)
1.57	Sodium carbonate (hexagonal)
1.54	Sodium carbonate (hexagonal)

Table 5
Composition of some agricultural lignocellulosic biomass [12].

Biomass	Composition (%, dry6 basis)		
	Cellulose	Hemicellulose	Lignin
Corn fiber	15	35	8
Corn cob	45	35	15
Corn stover	40	25	17
Rice straw	35	25	12
Wheat straw	30	50	20
Sugarcane bagasse	40	24	25
Switch grass	45	30	12
Coastal Bermuda grass	25	35	6

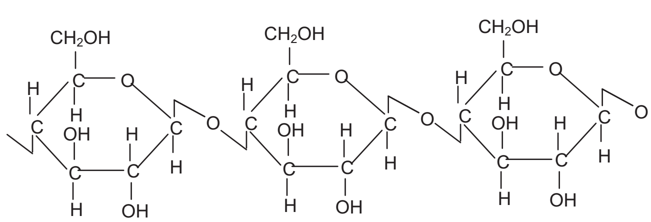


Fig. 2. Structure of cellulose.

cellulose: I–IV [17]. The natural form of cellulose known as cellulose I or native cellulose appears to be the most abundant form [18]. Cellulose II is generally obtained by regeneration of cellulose from solution or by mercerization. This allomorph is known as "regenerated" cellulose. The transition from cellulose I to cellulose II is not reversible implying that cellulose II is a stable form compared with the metastable cellulose I. Treatment with liquid ammonia or with certain amines such as ethylene diamine (EDA) allows the preparation of cellulose III either from cellulose I, which leads to the form cellulose III I or from cellulose II, which leads to the form III II. Cellulose III treated at a high temperature in glycerol is transformed into cellulose IV [19].

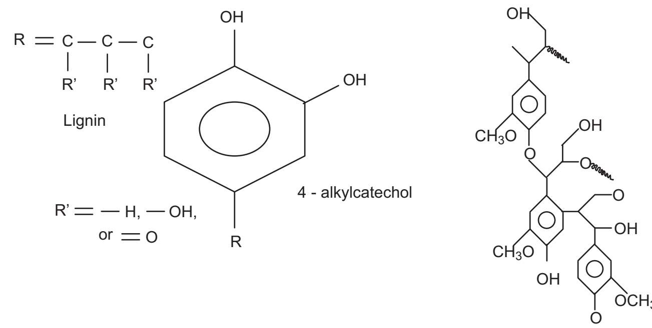


Fig. 3. Structure of lignin.

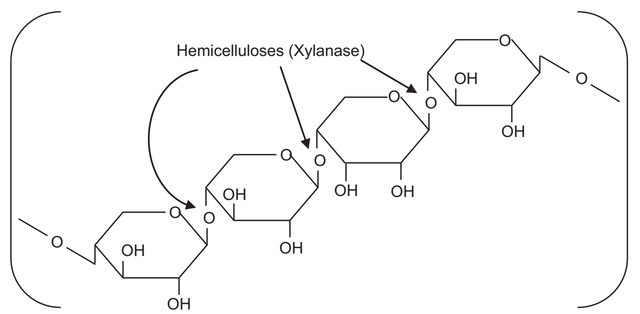


Fig. 4. Structure of hemicellulose.

Lignin is the principal aromatic component of wood. After cellulose, it is the most abundant renewable carbon source on earth. Around 40–50 million tons of lignin per annum is produced worldwide mostly as non-commercial waste product. The lignin molecule is a polymer with a degree of polymerization (DP) of 450–550, formed by the free radical, oxidative condensation of the three monomers, coniferyl alcohol, sinapyl alcohol and coumaryl alcohol [20]. Lignin is a complex polymer of phenyl propane units, which are cross-linked to each other with a variety of different chemical bonds as seen in Fig. 3. Lignin resists attack by most microorganisms. Lignin is nature's cement along with hemicellulose which exploits the strength of cellulose while conferring flexibility.

Hemicellulose structure, shown in Fig. 4, are heterogeneous polymers of pentoses (xylose, arabinose), hexoses (mannose, glucose, galactose), and sugar acids. Unlike cellulose, hemicelluloses are not chemically homogeneous. Hardwood hemicelluloses contain mostly xylans whereas softwood hemicelluloses contain mostly glucomannans. Besides xylose, xylans may contain arabinose, glucuronic acid or its 4-O-methyl ether and acetic, ferulic, and *p*-coumaric acids. The frequency and composition of branches are dependent on the source of xylan.

1.5. Challenges in the conversion of P. hysterophorus to ethanol

Since it is an obnoxious weed hence it's handling causes several health effect to human being.

Parthenium contains parthenin and some other toxcins which may hinder the growth of microorganism and therefore a reduced fermentation. It may also interfere with the process of enzymatic saccharification. Hence, removal/destruction of parthenin is a great challenge in the process of bioconversion to ethanol. Researchers should take utmost care in handling the raw material to avoid health hazards and also in the removal of toxins for higher yield of ethanol.

2. Process for conversion of *P. hysterophorus* to ethanol

The process of conversion starts with collection of fresh parthenium plants followed by drying, sizing and grinding. The biomass is then subjected to pretreatment via acid/alkali hydrolysis to achieve total available sugar and also to separate lignin. After pretreatment, simultaneous saccharification and fermentation steps are followed to produce extracellular endo-glucanase and β-glucosidase that are able to reduce cellulose and hemicelluloses to 6-carbon and 5-carbon sugars and subsequent fermentation to ethanol. The conversion of lignocellulosic parthenium to ethanol can be explained through simple flow diagram as shown in Fig. 5.

3. Materials and methods

Fresh parthenium plants are collected from the field and subjected to the following steps for experiments.

3.1. Preparation of *P. hysterophorus*

P. hysterophorus with long stem may be considered for the experiment which may be collected from field. It needs to be washed thoroughly for several times with tap water to remove adhering dirt, which needs to be chopped into small pieces (1–2 cm), dried in a hot air oven at 105 °C for 6 h and finally blended to small particles (1–2 mm). The dried material needs to be stored in airtight containers at room temperature until used.

3.2. Pretreatment

The effect of pretreatment of lignocellulosic materials has been recognized for a long time. The purpose of the pretreatment is to remove lignin, reduce cellulose crystallinity and increase the porosity of the materials. Pretreatment must meet the following requirements:

- Improve the formation of sugars or the ability to subsequently form sugars by enzymatic hydrolysis;
- Avoid the degradation or loss of carbohydrate;
- Avoid the formation of byproducts inhibitory to the subsequent hydrolysis and fermentation processes;
- Be cost-effective.

Physical, physico-chemical, chemical, and biological processes have been used for pretreatment of lignocellulosic materials. The following processes can be used for pretreatment of *P. hysterophorus*.

3.2.1. Mechanical comminution

Mechanically based pretreatment technologies are aimed at reducing the size of lignocellulosic wastes to facilitate subsequent treatments. Reduction of biomass size below #20 sieves shows the best mechanical performance [21]. Studies show that mechanical pretreatment technologies increase the digestibility of cellulose and hemicellulose in the *P. hysterophorus*. The use of mechanical chopping [21], hammer milling [22,23], grind milling [24], roll milling [25], vibratory milling [26] and ball milling [27] have been proved to be successful as a low cost pretreatment strategy.

Vibratory ball milling can be more effective in breaking down the cellulose crystallinity of certain lignocellulosic biomass and improving the digestibility of the biomass than ordinary ball milling. It results in improved digestibility of cellulose and hemicellulose to glucan and xylan respectively; it further enhance enzymatic digestibility with lower enzyme loads. Mechanical pretreatment also results in substantial lignin depolymerization via the cleavage of uncondensed-aryl ether linkages [27].

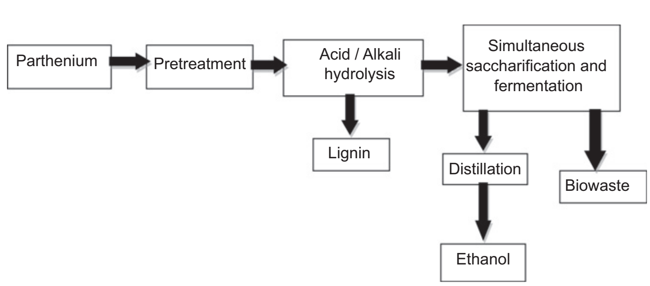


Fig. 5. Process flow diagram of lignocellulosic ethanol.

3.2.2. Acid hydrolysis

Lignocellulose of *P. hysterophorus* can be hydrolyzed using different acids to produce xylose, arabinose, glucose and acetic acid by cleavage of the β-1, 4 linkages of glucose or xylose monomers, acetyl groups and other products in cellulose and hemicellulose components of *P. hysterophorus*. The overall fermentable sugar available by acid hydrolysis may be 90% of the theoretical value of the sugar present in cellulosic biomass [28,29]. Dilute acid processes are conducted under high temperatures of 120–200 °C, high pressures of 15–75 psi and have reaction times in the range of 30 min to 2 h in continuous processes [30,31]. In dilute acid hydrolysis, the feedstock needs to be pressed before pretreatment either in dewatering presses or in compression screw feeders. The concentrated acid processes may be successful, for producing higher yields of sugar. These processes needs to be conducted with 60–90% sulfuric acid, mild temperatures and moderate pressures which is to be created by pumping materials from one vessel to another vessel for effective hydrolysis. The primary advantage of the concentrated acid process may be the high sugar recovery efficiency which can be of the order $>90\%$ for both xylose and glucose sugars [30,32]. Acid hydrolysis processes have several disadvantages due to formation of toxic compounds such as furfural, hydroxyl-methyl furfural, acetic acid, formic acid, levulinic acid etc., which inhibit the fermentation.

3.2.3. Organosolv process

Organic acids such as oxalic, acetylsalicylic and salicylic acid can be used as catalysts in the organosolv process whereby an organic or aqueous organic solvent mixture with inorganic acids (HCl or H_2SO_4) are used to break the internal lignin and hemicellulose bonds. The organic solvents for the process may include methanol, ethanol, acetone, ethylene glycol, triethylene glycol and tetrahydrofurfuryl alcohol [32,33]. Solvents used in the process needs to be drained from the reactor, evaporated, condensed and recycled to reduce the cost. Removals of solvents from the system are necessary because the solvents may cause inhibition to the growth of organisms, enzymatic hydrolysis and fermentation.

3.2.4. Enzymatic hydrolysis

Research indicates [34,35] that a chemical pretreatment followed by enzymatic hydrolysis increases the overall saccharification efficiency. The process of conversion of polysaccharides into fermentable sugars is called saccharification. It is a biochemical process in which hemicellulosic and cellulosic portions are converted into monomeric sugars and sugar acids. Various extracellular enzymes are required for cellulose hydrolysis and the hydrolyzed products are transported inside the cell for further catabolism. Table 6 represents various enzymes required for cellulose hydrolysis. Cellulose decomposition is initiated by a diverse

Table 6
Enzymes involved in the hydrolysis [13].

Enzyme	Mode of action
Endo-xylanse	Hydrolyzes β-1, 4-xylose linkages of the xylan back bone
Exo xylanase	Hydrolyzes β-1, 4-xylose linkages releasing xylobio
B-Xylosidase	Releases xylose from xylobiose and short Chain xylooligosaccharides
α-Arabinofuranosidase	Hydrolyzes terminal nonreducing α-arabinofuranose from Arabinoxylans
α-Glucuronidase	Releases glucurinic acid from glucuronoxylans
Acetylxylan	Hydrolyzes acetylester bonds in acetyl xylans
Ferulic acid esterase	Hydrolyzes feruloylester bonds in xylans
p-Coumaric acid esterase	Hydrolyzes *p*-coumaryl ester bonds in xylans

group of enzymes called cellulases namely β-1,4-endoglucanases, β-1,4-exoglucanases and β-1,4-glucosidases. Studies show that endoglucanases carry out cellulose mineralization which involves the loss of crystallinity of the structure followed by the depolymerization activity by exoglucanases leading to the production of linear chains of 2–3 glucose units called cellobiose and cellotriose. The cellobiose and cellotriose may enter into the cell where β-1,4-glucosidases are able to hydrolyze these to monomeric glucose units.

3.2.5. *Alkaline pretreatment*

Alkaline pretreatment is one of major chemical pretreatment technologies receiving numerous studies. It may employ various bases including sodium hydroxide [36–39], calcium hydroxide (lime) [40–42], potassium hydroxide [43], aqueous ammonia [44,45], ammonia hydroxide [46] and sodium hydroxide in combination with hydrogen peroxide or others [47–49].

Alkaline pretreatment is basically a delignification process in which a significant amount of hemicellulose may be solubilized. The action mechanism is believed to be saponification of intermolecular ester bonds cross linking xylan hemicelluloses and other components, for example, lignin and other hemicellulose alkaline pretreatment of lignocellulosic materials causes swelling, leading to decreased delignification process and crystallinity, increased internal surface area, disruption of the lignin structure and separation of structural linkages between lignin and carbohydrates [50]. Lime pretreatment at ambient conditions for up to 192 h [51] may enhance the enzyme digestibility using alkali chemicals [NaOH, $Ca(OH)_2$ and KOH] to pretreat rice straw in 24 h at 25 °C. Researchers found that NaOH (6% chemical loading, g/g dry rice straw) was the best alkali chemical to achieve 85% increase of glucose yield by enzymatic hydrolysis. Aqueous ammonia may also be used as a common alkali for alkaline pretreatment. In comparison with other pretreatment technologies, alkali pretreatment usually conducted under lower temperatures and pressures, even ambient conditions. Pretreatment time, however, are recorded in terms of hours or days which are much longer than other pretreatment processes. A significant disadvantage of alkaline pretreatment is the conversion of alkali into irrecoverable salts and/or the incorporation of salts into the biomass during the pretreatment reactions so that the treatment of a large amount of salts becomes a challenging issue for alkaline pretreatment.

3.3. *Biological process*

Biological treatment involves the use of whole organisms or enzymes in pretreatment of lignocellulose. Both fungi and bacteria may be used for biotreatment. Commercial preparations of fungal and bacterial hydrolytic and oxidative enzymes may also be used instead of these microorganisms. Fungal pretreatment of agricultural residues may be used for the improvement of digestibility [52]. White-, brown- and soft-rot fungi are generally used to degrade lignin and hemicellulose in plant materials whereby brown-rots mainly attack cellulose, while white and soft rots attack both cellulose and lignin. White-rot fungi are the most effective basidiomycetes for biological pretreatment of lignocellulosic materials [32]. Recent studies have shown that *Aspergillus terreus* [53]; *Trichoderma* sp. [54]; *Cyathus stercoreus* [55]; *Lentinus squarrosulus* [56]; *Penicillium camemberti* [57] grown at 25–35 °C for 3–22 days resulted to 45–75% and 65–80% hemicellulose and lignin degradation respectively. Recombinant strains of *Saccharomyces cerevisiae* can be genetically engineered to carry out simultaneous saccharification and fermentation (SSF) to produce extracellular endo-glucanase and β-glucosidase that are able to ferment cellulose and hemicellulose to 6-carbon and 5-carbon sugars and subsequent fermentation to ethanol [58–62]. After pretreatment the substrate is ready for fermentation.

3.4. *Fermentation*

Fermentation is the process of conversion of sugar into alcohol. To obtain ethanol from pentose and hexose sugar from hemicelluloses and cellulose the following steps are to be followed.

3.4.1. *Preparation of fermentation medium*

Ten grams of neopeptone needs to be added to the over limed hydrolysate and the pH of the solution may be adjusted to 5.6. This solution needs to be placed in 2 L Erlenmeyer flask filled up with distilled water up to 1 L and autoclaved at 121 °C, 15 lb for 15 min.

3.4.2. *Fermentation of P. hysterophorus hydrolysate to alcohol*

Two full plates of *Candida shehatae* on SXA medium needs to be inoculated into the fermentation medium and may be further incubated at 30 °C for 3 weeks. Samples needs to be aliquoted at different time intervals and assayed for ethanol content. For comparison Sabouraud Dextrose Broth (SDB) and Sabouraud Xylose Broth (SXB) (containing 20 g dextrose and xylose respectively) may be used as control media.

4. Kinetic model for hemicellulose hydrolysis

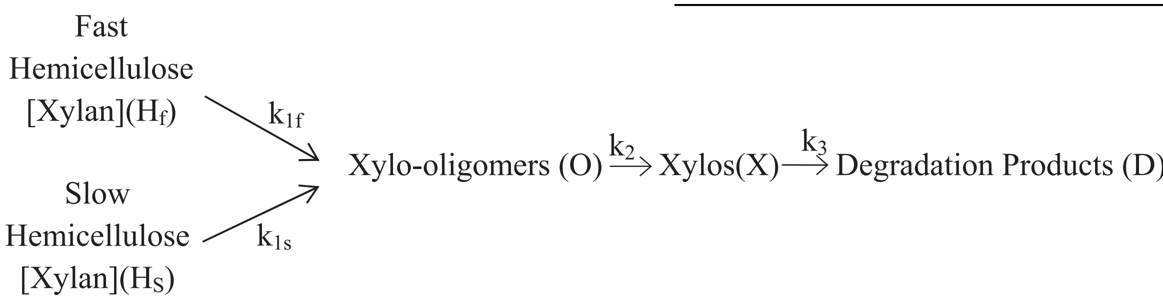
In mid-1940s, Saeman [63] modeled the first-order homogeneous kinetics of wood cellulose saccharification and it became the pioneer model for describing sugar degradation followed by hemicellulose hydrolysis.

Monophasic hydrolysis of hemicellulose is described [2] as

$$\text{Hemicellulose [Xylan] } [H] \xrightarrow{k_1} \text{Xylo-oligomers (O)} \xrightarrow{k_2} \text{Xylos(X)} \xrightarrow{k_3} \text{DegradationProducts(D)}$$

In the mid-1950s, this model was modified by assuming that hemicelluloses was composed of two distinct fractions, one that is relatively easy to hydrolyze and the other more difficult.

The biphasic hydrolysis of hemicellulose [2] is described as



(2)

The variation of components such as hemicellulose, xylose monomer, xylo-oligomers and degradation products at any instant of time during the hydrolysis reactions can be theoretically determined through solution of a given set of differential equations for hemicellulose hydrolysis [2] as shown in Table 7.

Another approach considering severity model, R_0, at operating conditions such as time and temperature is applied to the following unique expression for water-only hydrolysis [64]:

$$R_0 = t\,\exp([T_H - T_R]/14.75) \quad (1)$$

where t is the reaction time in minutes; T_H is hydrolysis temperature and T_R is the reference temperature which is most often 100 °C. When acid is used, a combined severity parameter, CS that depends on the effect of added acid catalyst during organosolv treatment, has been applied [65].

$$\log CS = \log R_0 - \mathrm{pH} \quad (2)$$

When Eq. (1) is substituted into this expression, the following relationship results:

$$CS = t(\mathrm{H}^+)\exp([T_H - T_R]/14.75) \quad (3)$$

H^+ indicates the concentration of hydrogen ion. Most of the literature data on severity reports only weight percent acid addition but not pH. The expression is further modified [66] by assuming that the hydrogen ion concentration is proportional to the percent acid as below:

$$(\mathrm{H}^+)\alpha nA \quad (4)$$

where A is the acid concentration in weight percent and n is the constant of proportionality and is close to 10. This is comparable with many of the Saeman-based models [63] applied to hemicelluloses hydrolysis. Substituting Eq. (4) into Eq. (3) and assuming the proportionality constant $n=10$ gives a result, we termed the modified severity parameter, M_0:

$$M_0 = t \cdot 10A\,\exp([T_H = T_R]/14.75) \quad (5)$$

Although the value of the modified severity parameter, M_0 defined by Eq. (5) does not matches exactly with the CS defined by Eq. (3), but it helps to correlate different literature data that only provides weight percent acid addition and not pH.

4.1. Depolymerization of hemicellulose

Consider the breaking of one bond of a polymer, N, composed of n monomer units to form two new molecules, e.g., j & $n-j$:

$$N_n \rightarrow N_j + N_{n-j} \quad (6)$$

Subsequently, these products can degrade to produce new molecules N_k, N_i, N_{j-i}, N_{n-j-k}, as follows:

$$N_{n-j} \rightarrow N_k + N_{n-j-k} \quad (7)$$

$$N_j \rightarrow N_j + N_{j-i} \quad (8)$$

If it is assumed that all the bonds linking monomer units have the same probability of being broken, the rate of change in concentration of any j-mer can be expressed by the following differential equation:

$$\frac{dN_j}{dt} = 2K_h \sum\nolimits_{i=j+1}^{n} N_j - K(j-1)N_j \quad (9)$$

where K_h is the hydrolysis rate constant and is assumed to be the same irrespective of chain length. The first term on the right hand side is the rate of creation of j-mers from the scission of the molecules larger than a j-mer (there are two scission events that results the identical products) and the second term is the rate at which existing j-mers disappear when any of the $(j-1)$ bonds present are broken. If this expression is extended to the longest polymer chain of length n that can only be broken but not formed, the rate of change in its concentration can be expressed [66] as follows:

$$\frac{dN}{dt} = -K_h(n-1)N_n \quad (10)$$

5. Discussion

The results of depolymerization for a hypothetical pentamer can be predicted through solution [66] of the governing Eqs. (6)–(10) through simple integration technique. Fig. 6 shows how the series of reactions interplay during the depolymerization of hemicellulose. The same can be successfully used to predict the yield of xylose when parthenium feedstock is hydrolyzed with dilute sulfuric acid.

The reported work on the yield of ethanol from *P. hysterophorus* is limited. However, the production yields of ethanol obtained from the acid hydrolysis of different biomass has been studied. Some results[62] revealed that using the sulfuric acid hydrolysis following the bioconversion by *C. shehatae* yielded ethanol with the maximum content of 1.01 g l^{-1}, the maximum yield coefficient of 0.19 g g^{-1} and the productivity of 0.008 g l^{-1} h^{-1}. These values are well comparable to those obtained from the phenol-tolerant strain of xylose fermenting bacterium [57]. This report herein showed

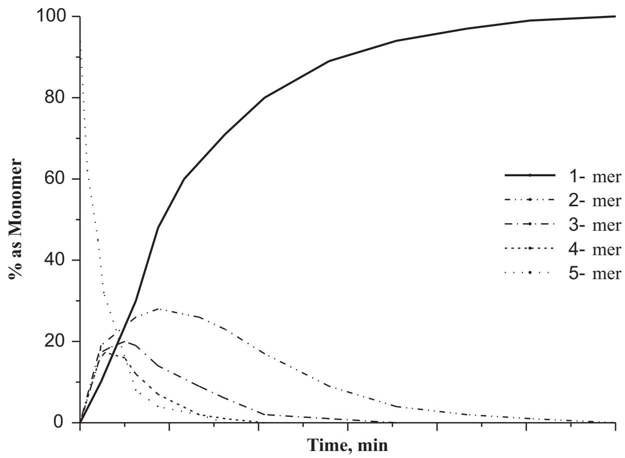


Fig. 6. Distribution curves for depolymerization of a hypothetical pentamer.

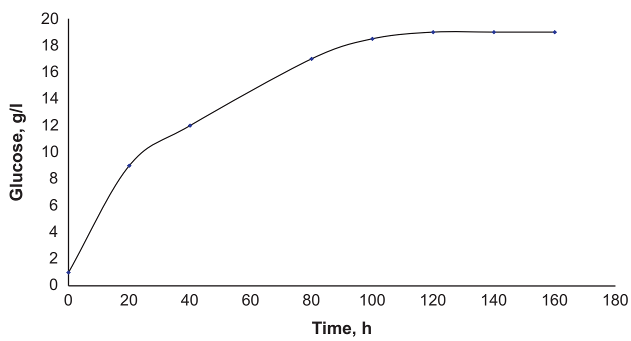


Fig. 7. Glucose yield.

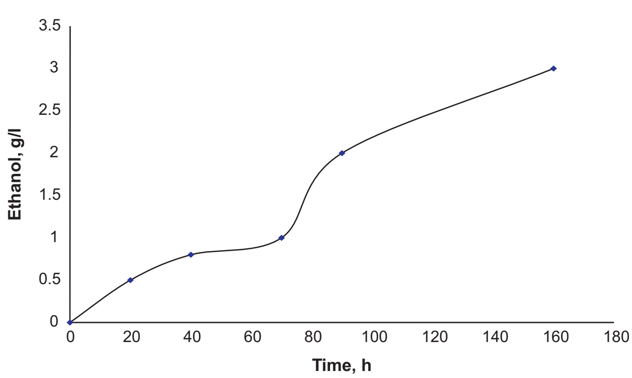


Fig. 8. Ethanol yield.

only 1.8 fold lesser in the coefficient yield than those obtained from using the fully-equipped fermentor. Therefore, further maximizations on the development of a versatile tool for ethanol production can be taken up for further research. There is a high feasibility of using an appropriate technology (acid hydrolysis and yeast fermentation) for the bioconversion from parthenium to ethanol. It is generally found that on an average, 1 kg of cellulose yield 1.1 kg of glucose and 1 kg of cellulose yield 0.56 kg of ethanol [67]. The glucose yield and ethanol yield from the process is shown in Figs. 7 and 8 [68].

6. Conclusion

Since *P. hysterophorus*, is threatening the biodiversity and human health in several areas throughout the world several researchers have documented the allelopathic effect of this weed. There is a need to encourage the research on the utilization of the weed. However, several studies proposed that Parthenium can be used as a green manure, compost, nanomedicines, biopesticide, agent for bio remediation for toxic metal and dyes etc. Parthenium, being a lignocellulosic biomass as well as wasteland weed, can be utilized in the process of lignocellulosic conversion into ethanol through either pretreatment, hydrolysis, simultaneous saccharification and fermentation or directly by auto hydrolysis and fermentation to produce eco-friendly green fuel which may offer very low cost inputs for ethanol production. The most challenging phase of this process is pretreatment, as it determines the volume of substrate for ethanol production. Pretreatment process like autohydrolysis is very important for the removal or destruction of perthenin which may have inhibitory effect on the action of enzymes added or the functioning of the yeasts. Research work needs to be carried out further in these regard and special emphasis may be given to develop an entire process with inputs of biological origin to get green ethanol from this obnoxious weed. This review work will stimulate the researchers to adopt a suitable kinetic model to study the reaction mechanism for hydrolysis of *Parthenium hystrophorus* L. and optimization of different parameters like temperature, time, acid and alkali concentrations, etc. during pretreatment process for achieving higher yield of ethanol.

Table 7
Governing equations for analysis of hemicellulose hydrolysis [2].

Monophasic hydrolysis	Biphasic hydrolysis
$\frac{dH}{dt}=-k_1[H]$	$\frac{dH_f}{dt}=-k_{1f}[H_f]$
$\frac{dO}{dt}=k_1[H]-k_2[O]$	$\frac{dH_s}{dt}=-k_{1s}[H_s]$
$\frac{dX}{dt}=k_2[O]-k_3[X]$	$\frac{dO}{dt}=k_{1f}[H_f]+k_{1s}[H_s]-k_2[O]$
$\frac{dD}{dt}=k_3[X]$	$\frac{dX}{dt}=k_2[O]-k_3[X]$
	$\frac{dD}{dt}=k_3[X]$

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